CHAPTER 2

Evidence for Nonthermal Electromagnetic Bioeffects: Potential Health Risks in Evolving Low-Frequency & Microwave Environments

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2.1 INTRODUCTION

In our solar system, the natural electromagnetic environment varies greatly from planet to planet. In the case of the planet Earth, a semiliquid ferromagnetic core generates a major and slowly migrating *static* geomagnetic field. Concurrently, there are much weaker natural *oscillating* low-frequency electromagnetic fields that arise from two major sources: in thunderstorm activity in equatorial zones of Central Africa and the Amazon basin; and in lesser degree, from solar magnetic storms in years of high activity in the 11-year solar sunspot cycle.

2.1.1 A Comparison of Natural and Man-Made Electromagnetic Environments

All life on earth has evolved in these fields. Defining them in physical terms permits direct comparison with far stronger man-made fields that have come to dominate all civilized environments in the past century. Energy in the *oscillating* natural fields is almost entirely in the extremely-low-frequency (ELF) spectrum, with peaks at frequencies between 8 and 32 Hz, the Schumann resonances (1957). Their electric components are around 0.01 V/m, with magnetic fields of 1-10 nanotesla. The earth's *static* magnetic field at 50 microtesla is 5000 times larger than the natural oscillations, but still substantially less than a wide range of daily human exposures to static and oscillating fields in domestic and occupational environments. Importantly, there is no significant level of natural energy at RF/microwave frequencies

Generation and distribution of electric power has spawned a vast and ever growing vista of new electronic devices and systems. They overwhelm the natural electromagnetic environment with more intense fields. They include oscillations far into the microwave spectrum, many octaves higher than the Schumann resonances. This growth of RF/microwave fields is further complicated by the advent of digital communication techniques. In many applications, these microwave fields,

oscillating billions of times per second, are systematically interrupted (pulsed) at low frequencies. This has raised important biological and biomedical questions, still incompletely answered, about possible tissue mechanisms in detection of amplitude- and pulse-modulation of RF/microwave fields (Adey, 1999).

2.1.2 Electric Power and Electronic Systems in Homes and Offices: Historical and Environmental Evidence

2.1.2.1 The Growth of Fields at Electric Power Frequenciess

A peak in childhood leukaemia at ages 2 through 4 emerged *de novo* in the 1920s in the United Kingdom and slightly later in the United States. Electrification in U.S. farms lagged behind urban areas until 1956. Using U.S. census data for 1930, 1940 and 1950, Milham and Osslander (2001) found that during 1928-1932, in states with above 75% of residences served by electricity, leukaemia mortality increased with age for single years 0-4, whereas states with electrification levels below 75%showed a decreasing trend with age ($P = 0.009$). During 1949-1951, all states showed a peak in leukaemia mortality at ages 2-4. At age 0-1, leukaemia mortality was not related to electrification levels. At ages 2-4, there was a 24% (95% CI, 8%-4!%) increase in leukaemia for a 10% increase in percentage of homes served by electricity. They conclude that the peak in the common childhood acute lymphoblastic leukaemia (ALL) may be attributable to electrification.

Design of modern office buildings has led to their electrification through one or more large distribution transformers that may be located in basement vaults, or in some cases, located on each floor of the building. Milham (1996) has examined cancer incidence in such a building over a 15-year period. A small cohort of 410 office workers (263 men and 147 women) were exposed to strong magnetic fields from three 12 kV transformers located beneath their ground-floor office. Cancers were detected in eight subjects over the 15-year period. There were indications of *cumulative risks*. Only one cancer was detected in 254 workers employed for less than 2 years, compared to seven cancers detected in 156 workers employed for 2 years or more ($P = 0.0057$; Fisher's exact test). An analysis of linear trend in cancer incidence, using average years employed as an exposure score, was positive ($P = 0.00337$), with an odds ratio of 15.1 in workers employed longer than 5 years. This positive correlation of cancer cases with duration of employment was seen for males and females separately and together ($P < 0.05$). For workers employed more than 2 years, the standardized cancer incidence ratio was 389 (95% confidence interval, 156-801).

None of these studies support tissue heating as an adequate model for the observed bioeffects.

2.1.2.2 The Appearance of Office Wireless Networks

In large modern offices, the electromagnetic environment has been further complicated by introduction of local-area-networks (LANs) for local telephonic and data transmission. Operating at gigahertz microwave frequencies, workers are

continuously exposed to fields from a plethora of sources located on each computer and on network hubs. Many of these sources utilise some form of low-frequency pulsed modulation. Their power output is typically in the low milliwatt to microwatt range - so low that significant heating of workers' tissues is improbable. Any bioeffects attributable to their operation strongly suggest *nonthermal* mechanisms of interaction, and raise further important questions about mechanisms mediating a cumulative dose from repeated, intermittent exposures, possibly over months and years.

Tissue components of environmental RF/microwave fields are consistent with two basic models. Sources close to the body surface produce *near-field exposures,*as with users of mobile phones. The emitted field is magnetically coupled directly from the antenna into the tissues. At increasing distances from the source, the human body progressively takes on properties of a radio antenna, with absorption of radiated energy determined by physical dimensions of the trunk and limbs. This is a *far-field exposure,* defined as fully developed at 30 wavelengths from the source. Permissible exposure limits (PELs) have rested on measurement of microwave field energy absorbed as heat, expressed as the *Specific Absorption Rate (SAR)* in W/kg.

The American National Standards Institute (1992) first recognised a tissue dose of 4.0 W/kg as a *thermal* (heating) tissue threshold likely to be associated with adverse effects and proposed an exposure limit in Controlled Environments (occupational) at 0.4 W/kg, thus creating a supposed "safety margin" of 10. For Uncontrolled Environments (civilian), a larger safety margin was set with a PEL 50 times lower at 0.08 W/kg. Since actual measurement of tissue SARs under environmental conditions is not a practical technique, PELs are typically expressed as a function of *incident field power density*, the amount of energy falling on a surface/unit area, and expressed in mW/cm^2. A simple yardstick relating incident field energy to its electric component relates an incident field of 1.0 mW/cm^2 to an electric field of 61V/m.

In the interim since formulation of these ANSI guidelines, there have been extensive reviews and proposed revisions. The U.S. government Interagency Radio Frequency Working Group (1999) has emphasised the need for revisions recognising nonthermal tissue sensitivities: "Studies continue to be published describing biological responses to nonthermal ELF-modulated RF radiation exposures that are not produced by CW (unmodulated) radiation." These studies have resulted in concern that "exposure guidelines based on thermal effects, and using information and concepts (time-averaged dosimetry, uncertainty factors) that mask any differences between intensity-modulated RF radiation exposure and CW exposure, do not directly address public exposures, and therefore may not adequately protect the public."

2.2 SENSITIVITIES TO NONTHERMAL STIMULI: TISSUE STRUCTURAL AND FUNCTIONAL IMPLICATIONS

2.2.1 Conductance Pathways in Multicellular Tissues

In its earliest forms, life on earth may have existed in the absence of cells, simply as a "soup" of unconstrained biomolecules at the surface of primitive oceans. It is a reasonable assumption that the first living organisms existed as single cells floating or swimming in these primordial seas. Concepts of a cell emphasise the role of a bounding membrane, surrounding an organised interior that participates in the chemistry of processes essential for all terrestrial life. This enclosing membrane is the organism's window on the world around it.

For unicellular organisms that swim through large fluid volumes, the cell membrane is both a sensor and an effector. As a sensor, it detects altered chemistry in the surrounding fluid, and provides a pathway for inward signals generated on its surface by a wide variety of stimulating ions and molecules, including hormones, antibodies and neurotransmitters. These most elemental inward signals are susceptible to manipulation by a wide variety of naturals or imposed electromagnetic fields that may also pervade the pericellular field. As effectors, cell membranes may also transmit a variety of electrical and chemical signals across intervening intercellular fluid to neighbouring cells, thus creating a domain or ensemble of cells, often able to "whisper together" in a faint and private language. Experimental evidence suggests that these outward effector signals may also be sensitive to intrinsic and imposed electromagnetic fields.

Rather than being separated in a virtually limitless ocean, cellular aggregates that form tissues of higher animals are separated by narrow fluid channels that take on special importance in signalling from cell to cell. Biomolecules travel in these tiny "gutters", typically not more than 150 wide, to reach binding sites on cell membrane receptors. These gutters form the *intercellular space* (ICS). It is a preferred pathway for induced currents of intrinsic and environmental electromagnetic fields. Although it occupies only ~ 10% of the tissue cross-section, it carries at least 90% of any imposed or intrinsic current, directing it along cell membrane surfaces. Whereas the ICS may have a typical impedance of ~ 4-50 ohm cm^{-1}, transmembrane impedances are ~ 10^4 - 10^6 ohm cm^{-2}.

2.2.2 Structural and Functional Organization of the Extracellular Space

Spaces in the ICS are not simple saline filled channels. Numerous stranded protein molecules protrude into these spaces from the cell interior and form a *glycocalyx* with specialised receptor sites that sense chemical and electrical stimuli in surrounding fluid. Their amino sugar tips are highly negatively charged. They offer an anatomical substrate for the first detection of weak electrochemical oscillations in pericellular fluid, including field potentials arising in activity of adjoining cells, or as tissue components of environmental fields. Research in molecular biology has increasingly emphasised essentially direct communication between cells due to

their mutual proximity. Bands of *connexin* proteins form *gap-junctions* directly uniting adjoining cell membranes. Experimental evidence supports their role in intercellular signalling

2.2.3 Tissue Detection of Low Frequency Fields and RF/Microwave Fields Amplitude-Modulated at Low Frequencies: Structural and Functional Options

Differential bioeffects, to be discussed below, have been reported between certain nonthermal RF/microwave fields with low-frequency amplitude- or pulse-modulation when compared to exposures to unmodulated continuous wave (CW) fields at similar power levels. The findings suggest, but do not yet establish unequivocally, that this frequency dependence may be a system property in a sequence of molecular hierarchies beyond the first transductive step. If the concept of modulation frequency-dependence continues to gain support in further research, answers must be sought as to the manner of its detection.

For ELF fields, models based on joint static/oscillating magnetic fields have been hypothesised. They include ion cyclotron resonance (Liboff, 1992), where mono- and divalent cations, such as potassium and calcium, abundant in the cellular environment, may exhibit cyclotron resonance at ELF frequencies in the presence of ambient static fields of less than 100 μT, such as the geomagnetic field. Other models describing ELF frequency dependence have considered phase transitions (Lednev, 1991) and ion parametric resonance (Blackman *et al.*, 1994), but interpretation of this frequency dependence based on ion parametric resonance remains unclear (Adair, 1998).

For amplitude or pulse-modulated RF/microwave fields, there is the implication that some form of *envelope demodulation* occurs in tissue recognition of ELF modulation components, but the tissue may remain essentially transparent to the same signal presented as an unmodulated carrier wave (Adey, 1981, 1999). However, crucial questions remain unanswered. It is not known whether biological low frequency dependence is established at the transductive step in the first tissue detection of the field, or whether it resides at some higher level in an hierarchical sequence of signal coupling to the biological detection system (Engstrom, 1997). For ELF magnetic fields, experimental evidence points to a slow time scale in inhibition of tamoxifen's antiproliferative action in human breast cancer cells (Harland *et al.*, 1999).

It is a principle of radio physics that extraction of ELF modulation information from an amplitude-modulated signal requires a *nonlinear element* in the detection system. This required nonlinearity may involve a spatial component, such as differential conduction in certain directions along the signal path; or the path itself may exhibit nonlinearities with respect to such factors as spatial distribution of electric charges at fixed molecular sites (so called *fixed charges*); or conduction itself may involve a nonlinear quantum process, as in electron tunnelling across the transverse dimensions of the cell membrane.

These constraints impose a further essential condition for demodulation to occur in the multicellular tissues of living organisms. There must be a *site for*

demodulation to occur. Evidence supports a role for cell membranes to act in this way, based not only on their intrinsic structure, but also on their proximity to neighbouring cells in the typical organisation of tissues of the body. Typical tissue organisation meets the three criteria outlined above, but as cautionary note, does not allow calculation of possible detection efficiency. Direct neighbour-neighbour cellular interactions will invite our further consideration of properties of cellular ensembles or domains in determining tissue threshold sensitivities.

2.2.3.1 Directional Differences in Tissue Signal Paths

As already noted, the narrow gutters of the intercellular spaces offer preferred conduction pathways, with conductivity 10^2-10^4 higher through extracellular spaces than through cell membranes (Adey *et al.*, 1963). Thus, the intercellular spaces become preferred pathways for *conduction along (parallel to) cell membrane surfaces,* and will reflect the changing directions and cross sections of myriad channels. Although predominantly an ionic (resistive) conduction pathway, it may also exhibit reactive components, due to the presence of protein molecules in solution.

2.2.3.2 Nonlinearities Related to Electric Charge Distribution

A suggested basis for envelope demodulation at cell surfaces may reside in the intensely anionic charge distribution on strands of glycoprotein that protrude from the cell interior, forming the glycocalyx (Bawin and Adey, 1976; Adey, 1999). As already noted, they provide the structural basis for specific receptor sites and they attract a surrounding cationic atmosphere, composed largely of calcium and hydrogen ions. This charge separation creates a Debye layer. In models and experimental data from resin particles, Einolf and Carstensen (1971) concluded that this physical separation creates a large virtual surface capacitance, with dielectric constants as high as 10^6 at frequencies below 1 kHz. Displacement currents induced in this region by ELF modulation of an RF field may then result in demodulation.

2.2.3.3 Electron Tunnelling in Transmembrane Conduction: Nonlinearities in Space and Time

Experimental studies of transmembrane charge tunnelling by DeVault and Chance (1966) and their more recent theoretical development by Moser *et al.* (1992) offer an example of extreme functional nonlinearity within the cell membrane. Chance described temperature-independent millisecond electron transfer over a temperature range from 120 K to 4 K. Considering a cell membrane transverse dimension of 40, Moser *et al.* noted that a variation of 20 in the distance between donors and acceptors in a protein changes the electron transfer rate by 10^{12}-fold. Concurrently in the time domain, the electron transfer rate is pushed from seconds to days, or a 10-fold change in rate for a 1.7 change in distance.

2.2.3.4 Issues of Comparability Between Bioeffects of ELF Fields, ELF-Modulated RF Fields, and Unmodulated (CW) RF Fields

From the beginning of these studies in the 1970s, it was noted that there were similarities in responses of tissues and cultured cells to environmental fields that were either in the ELF spectrum, or were RF/microwave fields modulated at ELF frequencies. Available evidence has indicated similarities between certain cell ionic and biochemical responses to ELF fields, and to RF/microwave fields amplitude-modulated at these same ELF frequencies, suggesting that tissue demodulation of RF/microwave fields may be a critical determinant in ensuing biological responses.

These findings have been reviewed in detail elsewhere (Adey, 1997, 1999). They are briefly summarized here in experiments at progressively more complex levels in the hierarchies of cellular organization. Early studies described calcium efflux from brain tissue in response to ELF exposures (Bawin and Adey, 1976; Blackman *et al.*, 1985), and to ELF-modulated RF fields (Bawin and Adey, 1975; Blackman *et al.*, 1979, 1985; Dutta *et al.*, 1984). Calcium efflux from isolated brain subcellular particles (synaptosomes) with dimensions under 1.0 μm also exhibit an ELF modulation frequency-dependence in calcium efflux, responding to 16 Hz sinusoidal modulation, but not to 50 Hz modulation, nor to an unmodulated RF carrier (Lin-Liu and Adey, 1982). In the same and different cell culture lines, the growth regulating and stress responsive enzyme ornithine decarboxylase (ODC) responds to ELF fields (Byus *et al.*, 1988; Litovitz *et al.*, 1993) and to ELF-modulated RF fields (Byus *et al.*, 1987; Litovitz *et al.*, 1993; Penafiel *et al.*, 1996).

In more recent studies also related to cellular stress responses, Goodman and Blank and their colleagues have reported rapid, transitory induction of heat-shock proteins by microtesla level 60 Hz magnetic fields (Lin *et al.*, 1997). In human HL60 promyelocytic cells these exposures at normal growth temperatures activated heat shock factor1 and heat shock element binding, a sequence of events that mediates stress-induced transcription of the stress gene HSP70 and increased synthesis of the stress response protein hsp70kd. Thus, the events mediating the field-stimulated response appeared similar to those reported for other physiological stressors (hyperthermia, heavy metals, oxidative stress), suggesting to the authors a general mechanism of electromagnetic field interaction with cells. Their further studies have identified endogenous levels of c-*myc* protein as a contributor to the induction of HSP70 in response to magnetic field stimulation (Lin *et al.*, 1998), with the hypothesis that magnetic fields may interact directly with moving electrons in DNA (Blank and Goodman, 1999, 2001; Blank and Soo, 2001).

Immune responses of lymphocytes targeted against human lymphoma tumour cells (allogeneic cytotoxicity) are sensitive to both ELF exposures (Lyle *et al.*, 1988) and to ELF-modulated fields, but not to unmodulated fields (Lyle *et al.*, 1983).

Communication between brain cells is mediated by a spectrum of chemical substances that both excite and inhibit transaction and transmission of information between them. Cerebral amino acid neurotransmitter mechanisms (glutamate, GABA and taurine) are influenced by ELF fields (Kaczmarek and Adey, 1974; Bawin *et al.*, 1996), and also by ELF-modulated microwave fields, but not by

unmodulated fields. Kolomytkin *et al.* (1994) examined specific receptor binding of three neurotransmitters to rat brain synaptosomes exposed to either 880 or 915 MHz fields at maximum densities of 1.5 mW cm^{-2}. Binding to inhibitory gamma-aminobutyric acid (GABA) receptors decreased 30% at 16 pulses/sec, but was not significantly altered at higher or lower pulse frequencies. Conversely, 16 pulses/sec modulation significantly increased excitatory glutamate receptor binding. Binding to excitatory acetylcholine receptors increased 25% at 16 pulses/sec, with similar trends at higher and lower frequencies. Sensitivities of GABA and glutamate receptors persisted at field densities as low as 50 μW cm^{-2}.

A selective absence of responses to unmodulated (CW) RF/microwave fields reported in many of these earlier studies has focused attention on establishment of threshold sensitivities to CW field exposures. De Pomerai *et al.* (2000) has reported cellular stress responses in a nematode worm as a biosensor of prolonged CW microwave exposures at athermal levels. Tattersall *et al.* (2001) exposed slices of rat hippocampal cerebral tissue to 700 MHz CW fields for 5-15 minutes at extremely low SARs in the range 0.0016-0.0044W kg^{-1}. No detectable temperature changes (+/- 0.1C) were noted during 15minute exposures. At low field intensities, a 20% potentiation of electrically evoked population potentials occurred, but higher field intensities evoked either increased or decreased responses. The exposures reduced or abolished chemically induced spontaneous epileptiform activity. Bawin *et al.* (1996) also tested the rat hippocampal slice, using ELF magnetic fields. At 56 μT (0.35-3.5 nV mm^{-1}), magnetic fields destabilised rhythmic electrical oscillations via as yet unidentified nitric oxide mechanisms involving free radicals.

2.3 INITIAL TRANSDUCTION OF IMPOSED ELECTROMAGNETIC FIELDS AT NONTHERMAL ENERGY LEVELS

There is an initial dichotomy in possible modes of interaction of cells in tissue with environmental microwave fields. It is principally determined by the separation of responses attributed to tissue heating from those elicited by certain fields at levels where frank heating is not the basis of an observed interaction. Their interpretation and possible significance has required caution in both biological and biophysical perspectives. Many of these biological sensitivities run counter to accepted models of physiological thresholds based in equilibrium thermodynamics of kT thermal collision energies. In a physical perspective, the search also continues for biological systems compatible with a first transductive step in a range of functionally effective vibrational and electromagnetic stimuli that are orders of magnitude weaker than kT. Their occurrence invites hypotheses on directions of future research.

2.3.1 Cell Membranes as the Site of Initial Field Transductive Coupling

Collective evidence points to cell membrane receptors as the probable site of first tissue interactions with both ELF and microwave fields for many neurotransmitters (Kolomyttkin *et al.*, 1994), hormones (Liburdy, 1995; Ishido *et al.*, 2001), growth-regulating enzyme expression (Byus *et al.*, 1987; Chen *et al.*, 2000; Litovitz *et al.*, 1993; Penafiel *et al.*, 1997), and cancer-promoting chemicals (Cain *et al.*, 1993; Mevissen *et al.*, 1999). In none of these studies does tissue heating appear involved causally in the responses. Physicists and engineers have continued to offer microthermal, rather than athermal, models for these phenomena (Barnes, 1996; Astumian *et al.*, 1995), with views that exclude consideration of cooperative organisation and coherent charge states, but it is difficult to reconcile experimental evidence for factors such as modulation frequency-dependence and required duration of an amplitude-modulated signal to elicit a response (*coherence time*) (Litovitz *et al.*, 1993) with models based on the equilibrium dynamics of tissue heating.

2.3.2 Evidence for Role of Free Radicals in Electromagnetic Field Bioeffects

Examination of vibration modes in biomolecules, or portions of these molecules, (Illinger, 1981) has suggested that resonant microwave interactions with these molecules, or with portions of their structure, is unlikely at frequencies below higher gigahertz spectral regions. This has been confirmed in studies showing collision-broadened spectra, typical of a heating stimulus, as the first discernible response of many of these molecules in aqueous solutions to microwave exposures at frequencies below 10 GHz.

However, there is an important option for biomolecular interactions with static and oscillating magnetic fields through the medium of *free radicals* (see Adey, 1993, 1997 for summaries). Chemical bonds are magnetic bonds, formed between adjacent atoms through paired electrons having opposite spins and thus magnetically attracted. Breaking of chemical bonds is an essential step in virtually all chemical reactions, each atomic partner reclaiming its electron, and moving away as a free radical to seek another partner with an opposite electron spin. The brief life time of a free radical is about a nanosecond or less, before once again forming a *singlet pair* with a partner having an opposite spin, or for electrons with similar spins, having options to unite in three ways, forming *triplet pairs* (reviewed in Adey, 1999).

During this brief lifetime, imposed magnetic fields may delay the return to the singlet pair condition, thus influencing the *rate* and the *amount of product* of an on-going chemical reaction (McLauchlan, 1992). McLauchlan points out that this model predicts a potentially "enormous effect" on chemical reactions for static fields in the low mT range. For oscillating fields, the evidence is less clear on their possible role as direct mediators in detection of ELF frequency-dependent bioeffects. *Spin-mixing* of orbital electrons and nuclear spins in adjacent nuclei is a possible mechanism for biosensitivities at extremely low magnetic field levels,

but these interactions are multiple, complex and incompletely understood (McLauchlan and Steiner, 1991). The highest level of free radical sensitivity may reside in hyperfine-dependent singlet-triplet state mixing in radical pairs with a small number of hyperfine states that describe their coupling to nearby nuclei (Till *et al.*, 1998; Timmel *et al.*, 1998). Singlet-triplet interconversion would need to be sufficiently fast to occur before diffusion reduced the probability of radical re-encounter to negligible levels.

Lander (1997) has emphasised that we are at an early stage of understanding free radical signal transduction. "Future work may place free radical signalling beside classical intra- and intercellular messengers and uncover a woven fabric of communication that has evolved to yield exquisite specificity." A broadening perspective on actions of free radicals in all living systems emphasises a dual role: first as messengers and mediators in many key processes that regulate cell functions throughout life; and second, in the pathophysiology of *oxidative stress diseases*.

At cell membranes, free radicals may play an essential role in regulation of receptor specificity, but not necessarily through a "lock and key" mechanism. As an example, Lander cites the location of cysteine molecules on the surface of P21-*ras* proteins at cell membranes. They may act as selective targets for nitrogen and oxygen free radicals, thereby inducing covalent modifications; and thus setting the *redox potential* of this target protein molecule as the critical determinant for its highly specific interactions with antibodies, hormones, etc. Magnetochemistry studies have suggested a form of cooperative behaviour in populations of free radicals that remain *spin-correlated* after initial separation of a singlet pair (Grundler *et al.*, 1992). Magnetic fields at 1 and 60 Hz destabilise rhythmic oscillations in brain hippocampal slices at 56 μT (0.35 to 3.5 nV mm^{-1}, via as yet unidentified nitric oxide mechanisms involving free radicals (Bawin *et al.*, 1996). In a general biological context, these are some of the unanswered questions that limit free radical models as general descriptors of threshold events.

2.4 THE ROLE OF CELLULAR ENSEMBLES IN SETTING TISSUE THRESHOLDS FOR INTRINSIC AND ENVIRONMENTAL STIMULI

Our pursuit of mechanisms mediating tissue electromagnetic sensitivities at nonthermal levels raises questions about the relevance of observed thresholds in the sensory physiology of other modalities. By extrapolation, do these data suggest the need to explore collective properties of populations of cells in setting thresholds by forms of intercellular communication? Do cooperative processes yield one or more faint and private languages that allow ensembles of cells to "whisper together" in one or more faint and private languages? Do observed *tissue* sensory thresholds differ significantly from thresholds measured in single cells in isolation from their neighbours?

2.4.1 Evidence for Domain Functions as a General Biological Property in Tissues

Research in sensory physiology supports this concept that some threshold properties may reside in highly cooperative properties of populations of elements, rather than in a single detector (Adey, 1998). Seminal observations in the human auditory system point to a receptor vibrational displacement of 10^{-11} m, or approximately the diameter of a single hydrogen atom (Bialek, 1983; Bialek and Wit, 1984); human olfactory thresholds for musk occur at 10^{-13} M, with odorant molecules distributed over 240 mm^2 (Adey, 1959); and human detection of single photons of blue-green light occurs at energies of 2.5 eV (Hagins, 1979). In another context, pathogenic bacteria, long thought to operate independently, exhibit ensemble properties by communication through a system recognizing colony numbers as an essential step preceding release of toxins. These *quorum sensing* systems may control expression of virulence factors in the lungs of patients with cystic fibrosis (Erickson *et al.*, 2002).

2.4.1.1 Domain Properties in Systems of Excitable Cells

Bialek addressed the problem of the auditory receptor in quantum mechanical terms. He evaluated two distinct classes of quantum effects: a *macroquantum effect* typified by the ability of the sensory system to detect signals near the quantum limits to measurement; and a *microquantum effect*, in which "the dynamics of individual biological macromolecules depart from predictions of a semi-classical theory." Bialek concluded that quantum-limited sensitivity occurs in several biological systems, including displacements of sensory hair cells of the inner ear. Remarkably, quantum limits to detection are reached in the ear in spite of seemingly insurmountable levels of thermal noise.

To reach this quantum limit, these receptor cells must possess amplifiers with noise performance approaching limits set by the uncertainty principle. It is equally impressive that suppression of intrinsic thermal noise allows the ear to function as though close to $0^{\circ}K$. Again, this suggests system properties inherent in the detection sequence. These "perfect" amplifiers could not be described by any chemical kinetic model, nor by any quantum mechanical theory in which the random phase approximation is valid. The molecular dynamics of amplifiers in Bialek's models would require preservation of quantum mechanical coherence for times comparable to integration times of the detector. It is not known whether comparable mechanisms may determine electromagnetic sensitivities as a more general tissue property at cellular and subcellular levels.

Behavioural electrosensitivity in sharks and rays may be as low as 0.5 nV mm^{-1} for tissue components of electrical fields in the surrounding ocean (Kalmijn, 1971). These marine vertebrates sense these fields through specialized jelly-filled tubular receptors (ampullae of Lorenzini) up to 10 cm in length, located near the snout and opening on the skin surface through minute pores. Sensing nerve cells lie in the wall of this ampullary tube. In support of a cooperative model of organisation of these neurons, behavioural electrosensitivity in sharks and rays is

100 times below measurable thresholds of individual electroreceptor neurons (Valberg *et al.*, 1997).

2.4.1.2 Domain Properties in System of Non-Excitable Cells: Culture Dimensions and "Bystander" Effects

Jessup *et al.* (2000) have pioneered studies on the role of gravitational fields in determining trends towards either apoptosis (programmed cell death) or towards cell proliferation. Concurrently, they tested the physical configuration of cell cultures in their influence on these same trends. Based on a colorectal cancer cell line, they compared cells cultured in adherent monolayers with three-dimensional ("3D") cultures. Biochemical measures of apoptosis and cell proliferation were tested in (1) static cultures, (2) in cultures subjected to slow rotation, and (3) in cultures exposed to the microgravity of low earth-orbital space flight. Over the course of 6 days on earth, static 3D cultures displayed the highest rates of proliferation and lowest apoptosis. Rotation appeared to increase apoptosis and decrease proliferation, whereas static 3D cultures in either unit-gravity or microgravity had less apoptosis. Expression of the carcinoembryonic antigen (CEA) as a marker of cell differentiation was increased in microgravity.

For ionising radiation, the U.S. National Council on Radiation Protection (NCRP) has recommended that estimates of cancer risk be extrapolated from higher doses by using a linear, no-threshold model. This recommendation is based on the dogma that the DNA of the nucleus is the main target of radiation-induced genotoxicity and, as fewer cells are directly damaged, the deleterious effects of ionising radiation proportionally decline. Experimental evidence seriously challenges this concept (Zhou *et al.*, 2001). They used a precision microbeam of α-particles to target an exact fraction (either 100% or $\leq$ 20%) of the cells in a confluent cell population and irradiated their nuclei with exactly one α-particle each. The findings were consistent with non-hit cells contributing significantly to the response, designated *the bystander effect.* Indeed, irradiation of 10% of a confluent mammalian cell population with a single α particle resulted in a mutant yield similar to that observed when all the cells in the population were irradiated. Importantly, this effect was eliminated in cells pretreated with 1mM octanol, which inhibits intercellular communication mediated by gap-junction proteins. "The data imply that the relevant target for ionising radiation mutagenesis is larger than an individual cell."

2.5 CONCLUSIONS

Epidemiological studies have evauated ELF and radiofrequency fields as possible risk factors for human health, with historical evidence relating rising risks of such factors as progressive rural electrification, and more recently, to methods of electric power distribution and utilisation in commercial buildings Appropriate models describing these bioeffects are based in nonequilibrium thermodynamics, with nonlinear electrodynamics as an integral feature. Heating models, based in

equilibrium thermodynamics, fail to explain an impressive spectrum of observed electromagnetic bioeffects at nonthermal exposure levels. We face a new frontier of much greater significance.

In little more than a century, our biological vista has moved from organs to tissues, to cells, and most recently, to the molecules that form the exquisite fabric of living systems. We discern a biological organization based in physical processes at the atomic level, beyond the realm of chemical reactions between biomolecules. Much of this signalling within and between cells may be mediated by free radicals of the oxygen and nitrogen species. In their brief lifetimes, free radicals are sensitive to imposed magnetic fields, including microwave fields. Free radicals are involved in normal regulatory mechanisms in many tissues. Disordered free radical regulation is associated with oxidative stress diseases, including Parkinson's and Alzheimer's diseases, coronary heart disease and cancer.

Although incompletely understood, tissue free radical interactions with magnetic fields may extend to zero field levels. Emergent concepts of tissue thresholds to imposed and intrinsic magnetic fields address ensemble or domain functions of populations of cells, cooperatively "whispering together" in intercellular communication, and organized hierarchically at atomic and molecular levels.

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